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ABUNDANCE AND MATING BEHAVIOR OF ORIENTAL FRUIT FLIES (DIPTERA: TEPHRITIDAE) NEAR METHYLEUGENOL-BAITED TRAPS

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Abstract.—Methyleugenol-baited traps are used for Oriental fruit fly control through male annihilation, as well as for detection and monitoring of fly populations. However, if the males which come to these traps emit sex pheromones, attract females from the surrounding vegetation, and mate with them before being killed, then using such traps might in fact increase levels of fruit infestation. In the present study, we monitored fly abundance in an experimental orchard before, during, and after methyleugenol-baited trap deployment. We also recorded the numbers of flies recruited to the trees with and without traps, and quantified their sexual activity. The males attracted by methyleugenol in our experiments fed on the poisoned baits almost immediately upon their arrival, and did not attempt to emit pheromones or attempt copulations before entering the traps. No changes in female abundance in the vicinity of deployed traps were recorded. Because of their high specificity, low cost, and environmental safety, methyleugenol-baited traps might be a valuable tool for integrated management of Oriental fruit fly populations.

Key Words.—Insecta, *Bactrocera dorsalis,* behavioral control, methyleugenol, mating, parapheromone.

Oriental fruit fly, *Bactrocera dorsalis* (Hendel), is one of the most important insect pests of tropical horticulture, which causes direct damage to more than 150 species of fruits and vegetables (Christenson & Foote 1960, Haramoto & Bess 1970). Male (but not female) Oriental fruit flies are strongly attracted to methyleugenol (4-allyl-1,2-dimethoxybenzene) (Steiner 1952). Feeding on this compound significantly increases male mating success (Shelly & Dewire 1994). Methyleugenol-baited traps have been widely used for Oriental fruit fly control through male annihilation, as well as for detection and monitoring of fly populations (Cunningham 1989). Male annihilation using methyleugenol has been successful in eradicating Oriental fruit fly from several isolated geographic areas, including the Marianas Islands (Steiner et al. 1965), the Amami Islands (Ushio et al. 1982), and the Okinawa Islands (Koyama et al. 1984). It has also been used to uproot the small "bridgehead" populations of the papaya fruit fly, *Bactrocera papayae* Drew and Hancock, which invaded Queensland, Australia in 1995 (Lloyd et al. 1998, Meats 1998, Hadwen et al. 1998). However, eradication of the Oriental fruit fly in Hawaii is hindered by the existence of large well-established fly populations, often in hard-to-reach natural areas (Vargas et al. 1989, 1990). Because of the high economic and environmental costs of fruit fly eradication, the goals of fruit fly control programs in Hawaii are beginning to shift from complete annihilation towards suppression of fly populations below economic thresholds in selected fruit-growing areas (Mitchell & Saul 1990, Vargas et al. 2000).

With a more targeted approach to the Oriental fruit fly management, we should

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anticipate an increased use of methyleugenol-baited traps for smaller-scale fly control within individual orchards. Cunningham & Suda (1986) achieved a 48% reduction in fruit infestation within a 63 ha papaya orchard by using only 9 small fiberboard blocks saturated with a mixture of methyleugenol and malathion per 1 ha of orchard area. However, using methyleugenol-baited traps within even smaller orchards (e.g., several ha) presents a potential danger. Male Oriental fruit flies attract females by secreting airborne sex pheromones (Kobayashi et al. 1978). They also display specialized courtship behaviors, expressed as rapid wing fanning and rear leg movements over abdomen, probably in an attempt to facilitate pheromone dispersal in the air (Arakaki et al. 1984, Shelly & Kaneshiro 1991). Therefore, if males which come to traps emit sex pheromones, attract females from surrounding vegetation, and mate with them before being killed, the use of methyleugenol traps might in fact increase levels of fruit infestation in localized areas. In the present study, we performed a series of experiments to investigate the possibility of this occurring in small orchards in Hawaii.

Methods and Materials

Experimental Sites.—The study was conducted between 28 Apr. and 13 Jun., 1999 at two sites on the Kauai Agricultural Research Center (Kapaa, island of Kauai, Hawaii). The two sites were isolated from each other by approximately 700 m of predominantly non-host vegetation. Site 1 was a small orchard of 75 grapefruit (*Citrus paradisi* Macf.) and eight mandarin (*C. nobilis* Lour.) trees, and Site 2 was a cluster of two shaddock (*C. grandis* (L.)), two grapefruit, and one mandarin trees adjacent to a coffee grove. The grapefruit trees at Site 1 were fruiting throughout the study, while no fruit was observed on any of the trees at Site 2 after 15 May.

Trap Design.—The methyleugenol-baited traps used in our experiments consisted of white plastic buckets (20 cm high, 20 cm in diameter) with 4 entry holes (2.5 cm in diameter) each. A single cotton wick (15.5 cm in length, 1 cm in diameter) was soaked in 10 ml of methyleugenol and 5 ml of a 10% water suspension of malathion and hung by wire from the inner side of each bucket's lid. Buckets were hung on the trees approximately 1.6 m above the ground.

Experiment 1.—The objective of our first experiment was to determine if deploying methyleugenol-baited traps leads to an increase in fly numbers in the immediate surrounding area. Twelve 22.5 by 13.75 cm yellow plastic rectangles (Trécé Inc., Salinas, California) covered with Tanglefoot were placed on trees at random locations within Site 1 (1 rectangle per tree). On the 7th day after rectangle deployment, 1 methyleugenol-baited trap was placed on a tree in the middle of the orchard for another 6 days, and then removed and taken to the laboratory for examination. The rectangles were replaced and checked for flies every 2 days for 18 consecutive days (6 days before placing the methyleugenol-baited trap in the orchard, 6 days during which the trap was deployed, and 6 days after the trap was removed). The numbers of captured male and female flies were recorded. The location of each replacement rectangle was selected at random. The experiment was repeated three times. Since the data did not follow a normal distribution (Wilk-Shapiro test, $n = 216$, $W = 0.5538$, $P < 0.05$ for the number of males, and $W = 0.8680$, $P < 0.05$ for the number of females), they were transformed using rank transformations (Conover & Iman 1981). Transformed data were analyzed using a two-way ANOVA (Analytical Software 1996), with the period during which the flies were captured (e.g., before, during, or after trap deployment) and replicates treated as main effects. When interaction terms were statistically significant, we also analyzed treatment effects separately within each replicate using one-way ANOVA.

Experiment 2.—In our second experiment, we investigated if there was any increase in courtship or mating activity of Oriental fruit flies in the areas immediately adjacent to methyleugenol-baited traps. One such trap was placed on a tree within one of the two sites. The other site was used as a control. The number of male and female flies, number of calling males, and number of mating pairs were recorded on the tree with a trap, as well as on the 4 trees immediately surrounding it. Observations began approximately 1 hour before sunset and continued for 40 minutes, thus covering the period of peak mating activity reported for the Oriental fruit fly (Arakaki et al. 1984, Shelly & Kaneshiro 1991). Observations were repeated 10 times. Each site was used five times as a treatment, and five times as a control. The numbers of flies recruited to the sites with and without a trap were compared using two-sample Student t-tests. One-tailed Fisher exact test (Zar 1999) was used to test the null hypothesis that the sex ratio of flies encountered around the trap was not skewed towards males. Two-tailed Fisher exact tests were used to test the null hypotheses that the sex ratio of flies observed in the control site was not different from 1:1 (Analytical Software 1996).

Experiment 3.—The goal of the third experiment was to determine if Oriental fruit fly males responding to methyleugenol arrive directly to the source of its odor. Two neighboring fruit trees were selected at random for observations. The distance between the trees was approximately 6.18 m (SE = 0.74). The number of flies on these trees was visually observed immediately before the beginning of the experiment, and their number was recorded. A methyleugenol-baited trap was then placed on one tree. The number of flies on the trees with and without the trap was then counted every 15 min for 1 h. Flies located on the outer or inner trap walls were not included in the count. The experiment was repeated five times at each of the two sites. Since the data did not follow a normal distribution (Wilk-Shapiro test, $n = 100$, $W = 0.8301$, $P < 0.05$), they were transformed using rank transformations (Conover & Iman 1981). Transformed data were analyzed using a three-way ANOVA (Analytical Software 1996), with site, time since trap placement, and presence of the trap treated as main effects. Means and standard errors were calculated from the non-transformed data only.

We also estimated the period of time between male arrival in the vicinity of a methyleugenol-baited trap and subsequent entry into the trap. Since all the trees within sites 1 and 2 had large canopies, preventing us from following the movements of individual flies, a methyleugenol-baited trap was placed on a potted guava tree (1.75 m tall) located approximately 10 m from Site 2. Time between fly landing on the guava tree and its entering the trap was recorded for 10 flies. The observations were repeated 4 times, with the behavior of a total of 40 flies observed.

Results

Experiment 1.—Experimental results and ANOVA statistics are presented in Table 1. Placing a methyleugenol-baited trap in the orchard significantly reduced

Table 1. Abundance of male and female Oriental fruit flies within Site 1 before, during, and after deploying of a methyleugenol-baited trap.

	Mean no. of males				Mean no. of females			
	Replicate 1	Replicate 2	Replicate 3	Overall	Replicate 1	Replicate 2	Replicate 3	Overall
Before trap placement	17.13	11.96	3.79	10.96	5.96	6.83	7.79	6.86
	(2.48)	(1.71)	(0.48)	(1.20)	(0.83)	(0.98)	(1.11)	(0.56)
During trap placement	14.29	4.46	2.17	6.97	5.50	10.79	5.88	7.39
	(1.51)	(0.53)	(0.33)	(0.82)	(0.81)	(1.52)	(0.85)	(0.69)
After trap placement	15.33	4.88	3.46	7.89	3.88	8.63	7.13	6.54
	(5.44)	(0.73)	(0.45)	(1.91)	(0.75)	(1.19)	(1.13)	(0.63)
ANOVA								
D.f.	2, 69	2, 69	2, 69	2, 207	2, 69	2, 69	2, 69	2, 207
F	2.93	12.86	4.91	10.64	2.62	2.81	0.98	0.89
P	0.0584	0.0001	0.0102	0.0001	0.0782	0.0653	0.3809	0.4148

the number of Oriental fruit fly males captured by yellow rectangles. There was substantial variation in the extent of this influence as indicated by a highly significant interaction term (ANOVA, df = 4, 207, $F = 4.49$, $P = 0.0018$). However, the general trend towards a reduction in male numbers due to trap deployment remained consistent within each replication. However, when the male population in the orchard was the highest (Replicate 1), the difference in the number of males caught before, during, and after trap deployment was not statistically different.

Unlike the numbers of captured males, female captures were not affected by trap presence. There was a significant interaction between treatment and replication (ANOVA, df = 4, 207, $F = 2.74$, $P = 0.0294$), indicating different fluctuation patterns in the numbers of females captured during each of the replications. However, separate analysis of treatment effects within each replication still did not reveal any significant influence of deploying the methyleugenol-baited trap on the number of females captured by yellow rectangles.

Experiment 2.—On average, 5.0 flies (SE = 0.98) encountered around methyleugenol-baited traps were identified as males, and 2.6 flies (SE = 1.01) encountered around the traps were identified as females. At the control sites, approximately 3.0 flies (SE = 1.22) were identified as males, and approximately 2.7 flies (SE = 1.04) were identified as females. Sex ratio was significantly skewed towards predominance of males in the presence of the trap (Fisher exact test, $\chi^2 = 3.89$, df = 1, $P = 0.0352$), but did not differ from 1:1 in the absence of the trap (Fisher exact test, $\chi^2 = 0.08$, df = 1, $P = 0.8515$).

Calling behavior was recorded for an average of 1.6 males (SE = 0.43) within the treatment sites, and an average of 1.4 males (SE = 0.64) within the control sites. The difference between the treatment and control sites was not statistically significant (Student *t*-test, $t = -0.26$, df = 18, $P = 0.7970$). Only 4 mating pairs were recorded during the entire study, with three of them being observed on a single day. All the observed mating took place within the treatment sites. However, when we included both mating and calling males in our analysis, the difference between the treatment and the control sites still remained statistically insignificant (Student *t*-test, $t = -0.69$, df = 18, $P = 0.5001$). No statistically significant difference in the number of recruited females was recorded between the treatment and the control sites (Student t-test, $t = 0.07$, df = 18, $P = 0.9460$).

Experiment 3.—Experimental results are shown in Fig. 1. Overall, significantly more flies were encountered at Site 2 than at Site 1 (ANOVA, df = 1, 80, $F = 32.21$, $P = 0.0001$), with the number of flies being significantly affected both by the presence of a trap (ANOVA, df = 1, 80, $F = 210.56$, $P = 0.0001$), as well as by the time since trap placement on a tree (ANOVA, df = 4, 80, $F = 11.20$, $P = 0.0001$). Time trends were similar at both sites, as indicated by the absence of a statistically significant interaction between the site and the time (ANOVA, df = 4, 80, $F = 0.94$, $P = 0.4451$). At the same time, the interaction between the time and the trap was highly significant (ANOVA, df = 4, 80, $F = 12.07$, $P = 0.0001$), with a rapid increase in the fly numbers on the trees with a trap soon after trap placement, and virtually no change on the trees without a trap (Fig. 1). A three-way interaction among site, trap, and the time since the trap placement was not statistically significant (ANOVA, df = 4, 80, $F = 1.23$, $P = 0.3057$). On average, flies spent 66.37 seconds (SE = 10.05) on the tree before entering the trap.

Discussion

Results of the present study once again demonstrate that methyleugenol-baited traps are highly attractive to Oriental fruit fly males. However, we did not find any increase in fly sexual activity in the vicinity of the deployed traps. It appears that the majority of males which are attracted to the odor of methyleugenol approach directly the source of the odor. Arrival rate curves observed in the present study for the flies landing on the tree containing a trap are similar to the curves reported by Nishida & Vargas (1990) for flies landing on their methyleugenol-baited traps. The recruited males feed on methyleugenol almost immediately upon their arrival and do not attempt to attract females and copulate before entering the trap. Such behavior is not unexpected, since male Oriental fruit flies incorporate methyleugenol metabolites into their sex pheromone (Nishida et al. 1988), and mating success of males fed on methyleugenol is significantly higher than mating success of males deprived of this compound (Shelly & Dewire 1994). Therefore, it is probably adaptive for males to ingest methyleugenol before trying to attract a mate. Actually, 40–50% of males respond to methyleugenol baits even before reaching full sexual maturity (Wong et al. 1989). Since there was no significant increase in male calling around the traps, and by itself methyleugenol is not very attractive to females (Steiner 1952), it is not surprising that we also did not detect any changes in female numbers following the deployment of methyleugenol-baited traps.

From a pest management perspective, it appears that using male lures on small farms will not attract additional Oriental fruit fly females from the surrounding vegetation, nor will it facilitate mating among the resident flies. Therefore, our original concern that the use of methyleugenol traps might in fact increase the level of fruit infestation appears to be unjustified. Still, it is unlikely that male annihilation alone can provide a satisfactory level of Oriental fruit fly control on small farms. This species is highly polygamous (Cunningham 1989). Therefore, even very few surviving males will be able to fertilize a substantial number of females, and each female can produce over 1000 eggs (Vargas et al. 1984). How-

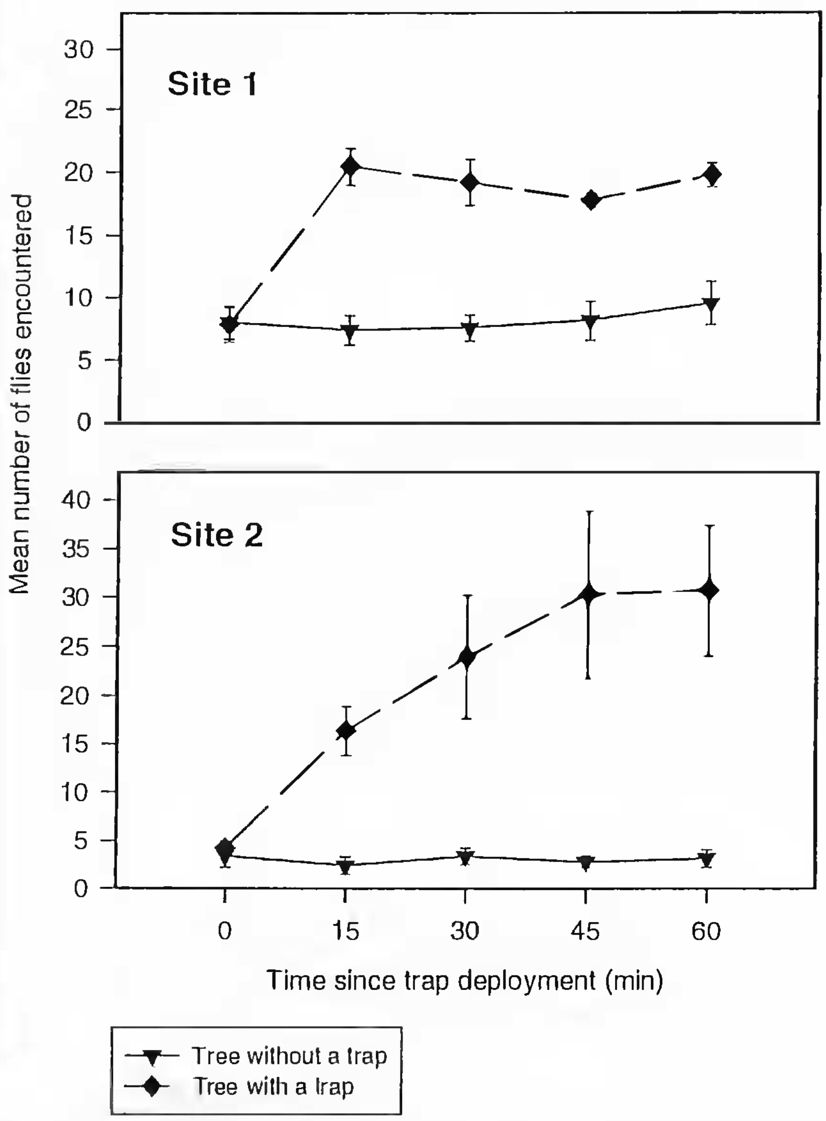

Figure 1. Effect of methyleugenol-baited traps on recruitment of Oriental fruit flies to fruit trees. The methyleugenol-baited trap consisted of a white plastic bucket with 4 entry holes and a single cotton wick soaked in methyleugenol-malathion mixture and hung by wire from the inner side of the bucket's lid. Error bars represent standard errors of measurement among 5 replications.

ever, low cost, high specificity, and environmental safety of methyleugenol-baited traps make them highly compatible with other management techniques, such as sterile male releases, biological control, bait sprays of environmentally friendly insecticides (e.g. spinosad), or visual lure-and-kill traps. Therefore, even though male lures alone are unlikely to solve the Oriental fruit fly problem, they might still be a valuable component of the future IPM systems.

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Literature Cited

Analytical Software. 1996. Statistix for Windows, version 1.0. Analytical Software, Tallahassee, Florida.

Arakaki, N., H. Kuba & H. Soemori. 1984. Mating behavior of the Oriental fruit fly, *Dacus dorsalis* Hendel (Diptera: Tephritidae). Appl. Ent. Zool., 19: 42–51.

Christenson, L. C. & B. H. Foote. 1960. Biology of fruit flies. Annu. Rev. Entomol., 5: 171–192.

Conover, W. J. & R. L. Iman. 1981. Rank transformations as a bridge between parametric and nonparametric statistics. Am. Stat., 35: 124–129.

Cunningham, R. T. 1989. Male annihilation. pp. 345-351. *In* Robinson, A. S. & G. Hooper (eds.). Fruit flies: their biology, natural enemies, and control. V. 3B. Elsevier, Amsterdam.

Cunningham, R. T. & D. Y. Suda. 1986. Male annihilation through mass-trapping of male flies with methyleugenol to reduce infestation of Oriental fruit fly (Diptera: Tephritidae) larvae in papaya. J. Econ. Entomol., 79: 1580–1582.

Hadwen, W. L., A. Small, R. L. Kitching & R. A. I. Drew. 1998. Potential suitability of North Queensland rain forest sites as habitat for the Asian papaya fruit fly, Bactrocera papayae Drew and Hancock (Diptera: Tephritidae). Austr. J. Entomol., 37: 219–227.

Haramoto, F. H. & H. A. Bess. 1970. Recent studies on the abundance of the oriental and Mediterreanean fruit flies and the status of their parasites. Proc. Hawaiian Entomol. Soc., 20: 551–566.

Kobayashi, R. M., K. Ohinata, D. L. Chamber & M. S. Fujimoto. 1978. Sex pheromones of the oriental fruit fly and melon fly: mating behavior, bioassay method, and attraction of females by live males and by suspected pheromone glands of males. Environ. Entomol., 7: 107–112.

Koyama, J., T. Teruya & K. Tanaka. 1984. Eradication of the Oriental fruit fly (Diptera: Tephritidae) from the Okinawa islands by a male annihilation method. J. Econ. Entomol., 77: 468–472.

Lloyd, A., P. Leach & R. Kopittke. 1998. Effects of exposure on chemical content and efficacy of male annihilation blocks used in the eradication of Bactrocera papayae in North Queensland. Gen. Appl. Entomol., 28: 1–8.

Meats, A. 1998. Cartesian methods of locating spot infestations of the papaya fruit fly Bactrocera papayae Drew and Hancock within the trapping grid at Mareeba, Queensland, Australia. Gen. Appl. Entomol., 28: 1–8.

Mitchell, W. C. & S. H. Saul. 1990. Current control methods for Mediterranean fruit fly, Ceratitis capitata, and their application in the USA. Rev. Agric. Entomol., 78: 923–940.

Nishida, R., K. H. Tan, M. Serit, N. H. Lajis, A. M. Sukari, S. Takahashi & H. Kukami. 1988. Accumulation of phenylpropanoids in the rectal glands of males of the Oriental fruit fly, *Dacus dorsalis*. Experientia, 44: 534–536.

Nishida, T. & R. I. Vargas. 1990. Arrival rates of the Oriental fruit fly, Dacus dorsalis Hendel (Diptera: Tephritidae) to methyl eugenol. Proc. Hawaiian Entomol. Soc., 30: 105–112.

Shelly, T. E. & K. Y. Kaneshiro. 1991. Lek behavior of the Oriental fruit fly, *Dacus dorsalis*, in Hawaii (Diptera: Tephritidae). J. Insect Behav., 4: 235–241.

Shelly, T. E. & A.-L. M. Dewire. 1994. Chemically mediated mating success in male Oriental fruit flies (Diptera: Tephritidae). Ann. Entomol. Soc. Am., 87: 375–382.

Steiner, L. F. 1952. Methyl eugenol as an attractant for Oriental fruit fly. J. Econ. Entomol., 45: 241–248.

Steiner, L. F., W. C. Mitchell, E. J. Harris, T. T. Kozuma & M. S. Fujimoto. 1965. Oriental fruit fly eradication by male annihilation. J. Econ. Entomol., 58: 961–964.

Ushio, S., K. Yoshioka, K. Nakasu & K. Waki. 1982. Eradication of the Oriental fruit fly from Amami Islands by male annihilation. Jpn. J. Appl. Entomol. Zool., 26:1–9.

Vargas, R. I., O. Miyashita & T. Nishida. 1984. Life history and demographic parameters of three laboratory-reared tephritids (Diptera: Tephritidae). Ann. Entomol. Soc. Am., 77: 651–656.

Vargas, R. I., J. D. Stark & T. Nishida. 1989. Abundance, distribution, and dispersion indices of the oriental fruit fly and melon fly (Diptera: Tephritidae) on Kauai, Hawaiian Islands. J. Econ. Entomol., 82: 1609–1615.

Vargas, R. I., J. D. Stark & T. Nishida. 1990. Population dynamics, habitat preference, and seasonal distribution patterns of oriental fruit fly and melon fly in an agricultural areas. Environ. Entomol., 19: 1820–1828.

Vargas, R. I., J. D. Stark, M. H. Kido, H. M. Ketter & L. C. Whitehead. 2000. Methyl eugenol and cue-lure traps for suppression of male Oriental fruit flies and melon flies (Diptera: Tephritidae) in Hawaii: effects of lure mixtures and weathering. J. Econ. Entomol., 93: 81–87.

Wong, T. T. W., D. O. McInnis & J. I. Nishimoto. 1989. Relationship of sexual maturation rate to response of Oriental fruit fly strains to methyl eugenol. J. Chem Ecol., 15: 1399–1495.

Zar, J. H. 1999. Biostatistical Analysis (4th ed.). Prentice Hall, Upper Saddle River, New Jersey.

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